PATENT COOPERATION TREATY

From the
INTERNATIONAL PRELIMINARY EXAMINING AUTHORITY

To:

DE CLERCQ, Ann
DE CLERCQ, BRANTS & PARTNERS
E. Gevaertdreef 10a
B-9830 Sint-Martens-Latem
BELGIQUE

NOTIFICATION OF TRANSMITTAL OF THE INTERNATIONAL PRELIMINARY EXAMINATION REPORT

(PCT Rule 71.1)

Date of mailing *(day/month/year)*	19.10.2004
Applicant's or agent's file reference: ALGO-002-PCT1	**IMPORTANT NOTIFICATION**

International application No.	International filing date *(day/month/year)*	Priority date *(day/month/year)*
PCT/EP 03/06049	10.06.2003	10.06.2002

Applicant: ALGONOMICS N.V. et al.

1. The applicant is hereby notified that this International Preliminary Examining Authority transmits herewith the international preliminary examination report and its annexes, if any, established on the international application.

2. A copy of the report and its annexes, if any, is being transmitted to the International Bureau for communication to all the elected Offices.

3. Where required by any of the elected Offices, the International Bureau will prepare an English translation of the report (but not of any annexes) and will transmit such translation to those Offices.

4. **REMINDER**

The applicant must enter the national phase before each elected Office by performing certain acts (filing translations and paying national fees) within 30 months from the priority date (or later in some Offices) (Article 39(1)) (see also the reminder sent by the International Bureau with Form PCT/IB/301).

Where a translation of the international application must be furnished to an elected Office, that translation must contain a translation of any annexes to the international preliminary examination report. It is the applicant's responsibility to prepare and furnish such translation directly to each elected Office concerned.

For further details on the applicable time limits and requirements of the elected Offices, see Volume II of the PCT Applicant's Guide.

The applicant's attention is drawn to Article 33(5), which provides that the criteria of novelty, inventive step and industrial applicability described in Article 33(2) to (4) merely serve the purposes of international preliminary examination and that "any Contracting State may apply additional or different criteria for the purposes of deciding whether, in that State, the claimed inventions is patentable or not" (see also Article 27(5)). Such additional criteria may relate, for example, to exemptions from patentability, requirements for enabling disclosure, clarity and support for the claims.

Name and mailing address of the international preliminary examining authority:

European Patent Office
D-80298 Munich
Tel. +49 89 2399 - 0 Tx: 523656 epmu d
Fax: +49 89 2399 - 4465

Authorized Officer

Schall, H

Tel. +49 89 2399-2647

PATENT COOPERATION TREATY

PCT

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

(PCT Article 36 and Rule 70)

Applicant's or agent's file reference	FOR FURTHER ACTION	See Notification of Transmittal of International Preliminary Examination Report (Form PCT/IPEA/416)
ALGO-002-PCT1		
International application No.	International filing date *(day/month/year)*	Priority date *(day/month/year)*
PCT/EP 03/06049	10.06.2003	10.06.2002
International Patent Classification (IPC) or both national classification and IPC		
G06F19/00		
Applicant		
ALGONOMICS N.V. et al.		

1. This international preliminary examination report has been prepared by this International Preliminary Examining Authority and is transmitted to the applicant according to Article 36.

2. This REPORT consists of a total of 5 sheets, including this cover sheet.

   ☒ This report is also accompanied by ANNEXES, i.e. sheets of the description, claims and/or drawings which have been amended and are the basis for this report and/or sheets containing rectifications made before this Authority (see Rule 70.16 and Section 607 of the Administrative Instructions under the PCT).

   These annexes consist of a total of 5 sheets.

3. This report contains indications relating to the following items:

   - I ☒ Basis of the opinion
   - II ☐ Priority
   - III ☐ Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
   - IV ☐ Lack of unity of invention
   - V ☒ Reasoned statement under Rule 66.2(a)(ii) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement
   - VI ☐ Certain documents cited
   - VII ☐ Certain defects in the international application
   - VIII ☐ Certain observations on the international application

Date of submission of the demand	Date of completion of this report
30.12.2003	19.10.2004
Name and mailing address of the international preliminary examining authority: European Patent Office D-80298 Munich Tel. +49 89 2399 - 0 Tx: 523656 epmu d Fax: +49 89 2399 - 4465	Authorized Officer Sisk, A Telephone No. +49 89 2399-6041

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

International application No. PCT/EP 03/06049

I. Basis of the report

1. With regard to the **elements** of the international application *(Replacement sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to this report since they do not contain amendments (Rules 70.16 and 70.17))*:

Description, Pages

1-46 as originally filed

Claims, Numbers

1-34 received on 06.10.2004 with letter of 06.10.2004

Drawings, Sheets

1-8 as originally filed

2. With regard to the **language**, all the elements marked above were available or furnished to this Authority in the language in which the international application was filed, unless otherwise indicated under this item.

These elements were available or furnished to this Authority in the following language: , which is:

- ☐ the language of a translation furnished for the purposes of the international search (under Rule 23.1(b)).
- ☐ the language of publication of the international application (under Rule 48.3(b)).
- ☐ the language of a translation furnished for the purposes of international preliminary examination (under Rule 55.2 and/or 55.3).

3. With regard to any **nucleotide and/or amino acid sequence** disclosed in the international application, the international preliminary examination was carried out on the basis of the sequence listing:

- ☐ contained in the international application in written form.
- ☐ filed together with the international application in computer readable form.
- ☐ furnished subsequently to this Authority in written form.
- ☐ furnished subsequently to this Authority in computer readable form.
- ☐ The statement that the subsequently furnished written sequence listing does not go beyond the disclosure in the international application as filed has been furnished.
- ☐ The statement that the information recorded in computer readable form is identical to the written sequence listing has been furnished.

4. The amendments have resulted in the cancellation of:

- ☐ the description, pages:
- ☒ the claims, Nos.: 35-46
- ☐ the drawings, sheets:

5. ☐ This report has been established as if (some of) the amendments had not been made, since they have been considered to go beyond the disclosure as filed (Rule 70.2(c)).

   (Any replacement sheet containing such amendments must be referred to under item 1 and annexed to this report.)

6. Additional observations, if necessary:

V. Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. Statement

Novelty (N)	Yes:	Claims	1-21,23-34
	No:	Claims	22
Inventive step (IS)	Yes:	Claims	1-21,23-34
	No:	Claims	22
Industrial applicability (IA)	Yes:	Claims	1-34
	No:	Claims	

2. Citations and explanations

   see separate sheet

Re Item I

Basis of the report

The basis of this report is the application as originally filed. Reference is made to the following documents cited in the international search report:

D1: WO 98/59244 A

D2: KNEGTEL RONALD M A ET AL: "Molecular docking to ensembles of protein structures." JOURNAL OF MOLECULAR BIOLOGY, vol. 266, no. 2, 1997, pages 424-440, XP002944096 ISSN: 0022-2836

a. The following clarity objections are made (Article 6 PCT):
(I) Claim 22 is unclear in its entirety. It also appears to be broader in scope than justified by the method of claim 1.
(ii) Claim 23 does not meet the requirements of Article 6 PCT, because it is not clear.
The accepted wording for a computer program claim is as follows:
"A computer program comprising the code means adapted to perform, when said program is run on a data processing system, all the method steps of 1 to x".

Re Item V

Reasoned statement with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

b. Document D1, which is considered to represent the closest prior art, discloses a system for predicting the binding affinity of a peptide to a MHC class molecule, by modelling all possible backbone structures for the peptide and then, for each backbone, modelling each possible side-chain conformation (see page 4, line 27 to page 5, line 2 and page 8, line 36 to page 9, line 9) and computing an affinity score for each peptide/molecule structure.
D1 differs from the system of claim 1 in that the following features are not present:
- the modelling of all peptide side chains, not just those at binding pockets
- calculating the potential energy of each complex
- calculating the conformational entropy of the complete ensemble.

c. Therefore, the subject-matter of claim 1 is new (Article 33(2) PCT).

d. With regards to inventive step, D1 has the problem that its binding affinity calculation is not optimally accurate when input data is scarce and does not

optimally separate between binding and non-binding peptides (the 'false-positive' problem).

e. The solution to this problem proposed in claim 1 of the present application is considered as involving an inventive step (Article 33(3) PCT) for the following reason. The system of claim 1 in the present application ensures a greater accuracy because:
- it models, for each peptide backbone structure, the side-chains of said peptide not just those that bind at a pocket
- it evaluates the potential energy of each modelled MHC/peptide complex
- it computes the term for conformational entropy from the ensemble of complexes.
The calculation of the conformation entropy for the complete ensemble provides greater accuracy of binding affinity.

f. Claims 2-21 are dependent on claim 1 and as such also meet the requirements of the PCT with respect to novelty and inventive step.

g. Claim 22 does not fulfill the requirements of the PCT with respect to novelty and inventive step (Article 33(3) PCT). Such a data representation is merely one of several straightforward possibilities from which the skilled person would select, in accordance with circumstances, without the exercise of inventive skill, in order to represent a peptide structure which binds to a MHC complex.

h. Claims 23 to 34 meet the requirements of the PCT with respect to novelty and inventive step.

CLAIMS (retyped)

1. A method for predicting the binding affinity of a peptide for a major histocompatibility (MHC) class I or class II molecule, comprising the following steps:

a) receiving a representation of a complete or partial three-dimensional structure of an MHC class I or class II molecule,

b) obtaining an ensemble of representations of peptide backbone structures of said peptide, said representations located within the binding site of said MHC molecule,

c) modeling for each peptide backbone structure of said ensemble in relation to said MHC molecule, at least the side-chains of said peptide, thereby obtaining an ensemble of modeled MHC/peptide complexes, and

d) evaluating the binding properties of said peptide for said MHC molecule, comprising at least two scoring elements:

d1) evaluating one or more components of the potential energy of each complex of the ensemble,

d2) evaluating the conformational entropy for the complete ensemble.

2. A method according to claim 1 wherein said representation of step (a) is obtained from one of the following:

- one or more experimentally determined structures obtained by for example X-ray crystallography, nuclear magnetic resonance spectroscopy, scanning microscopy, or
- one or more models derived from an experimentally determined structure, whereby said experimentally determined structure has a high sequence identity to said MHC molecule.

3. A method according to claim 1 or 2 wherein said representation of step (b) is generated by a computer modeling method, said method being able to generate multiple energetically favorable backbone configurations in relation to said MHC molecule.

4. A method according to claim 1 or 2 wherein said representation of step (b) is retrieved from a library of peptide structures pre-oriented in relation to said MHC molecule.

5. A method according to any of claims 1 to 4 wherein a complex within said ensemble of step (c) is obtained from a side-chain placement algorithm.

6. A method according to any of claims 1 to 5 wherein the side-chain placement of step (c) not only involves placing the side-chains of the peptide itself, but also involves placing at least one side-chain of said MHC molecule that are in contact with said peptide.

7. A method according to any of claims 1 to 6 wherein a complex within said ensemble of step (c) is obtained from a side-chain placement algorithm suited for global side-chain optimization.

8. A method according to any of claims 5 to 7 wherein the side-chain placement algorithm is a dead-end elimination (DEE) algorithm, characterized in that said DEE algorithm eliminates rotameric conformations on the basis of a mathematical criterion that allows the detection of conformations that are not compatible with the globally optimal conformation.

9. A method according to any of claims 5 to 7 wherein the side-chain placement algorithm is a FASTER algorithm, said algorithm being characterized by a repeated perturbation, relaxation and evaluation step.

10. A method according to any of claims 1 to 9 wherein the binding affinity of step (d) is represented by a single scoring value for the whole ensemble of MHC/peptide complexes, said scoring value comprising the sum of the conformational entropy for the complete ensemble of MHC/peptide complexes, and the average of the said energetical components of each of the complexes of said ensemble.

11. A method according to any of claims 1 to 10 wherein the binding affinity of step (d) is evaluated for the global complex, thereby accounting for interactions between pairs of residues from the peptide, the MHC molecule and both the peptide and the MHC molecule.

12. A method according to any of claims 1 to 11 wherein the entropical component reflects the overall conformational flexibility of the peptide.

13. A method according to any of claims 1 to 12 wherein the representations of said peptide contained in said library are derived from experimentally determined structures.

14. A method according to any of claims 1 to 12 wherein the representations of said peptide contained in said library are derived from computer-generated structures, said structures generated by said computer modeling method of claim 3.

15. A method according to any of claims 1 to 14 wherein said peptide comprises one or more non-naturally occurring amino acids.

16. The method according to any of claims 1 to 15 applied to multiple selected peptides by repeated application of said method for a single peptide.

17. The method of claim 16 wherein said multiple selected peptides are one or more putative immunogenic peptide fragments derived from a polypeptide of interest.

18. The method according to claims 16 to 17 further comprising the steps of

(a) inferring one or more putative immunogenic peptides that bind to said MHC molecule,

(b) optionally preparing one or more of said putative immunogenic peptides of said polypeptide of interest,

(c) optionally testing complexes of said one or more putative immunogenic peptides of said MHC molecule for an ability to be recognized by MHC cytotoxic T cells, and to thereby induce a cytotoxic T cell response to the epitope within the immunogenic peptide, and

(d) selecting said one or more putative immunogenic fragments comprising an MHC class I or class II binding site that induce an MHC class I or class II cytotoxic T cell response to the epitope.

19. The method according to any of claims 16 to 18 for producing an immunogenic peptide comprising an MHC class I or class II restricted T cell epitope that binds to an MHC class I or class II molecule and induces an MHC class I or II -restricted cytotoxic T cell response.

20. A method according to any of claims 1 to 19 wherein said MHC class I molecule comprises an HLA antigen selected from any of the HLA-A, HLA-B, HLA-C, HLA-E, HLA-F and HLA-G alleles.

21. A method according to any of claims 1 to 19 wherein said MHC class II molecule comprises an HLA antigen selected from any of the HLA-DR, HLA-DQ and HLA-DP gene products.

22. Data comprising

- representations of one or more peptide backbone structures, each peptide demonstrating an interaction with an MHC class I or class II molecule, and

- an indication of the MHC molecule associated with said representation.

23. A computer program comprising computing routines, stored on a computer readable medium for evaluating the binding affinity of a peptide for an MHC class I or class II molecule, said routines comprising:

- receiving an ensemble of representations of structures of the complex between said MHC molecule and said peptide,
- evaluating one or more components of the potential energy of each complex of the ensemble,
- evaluating the conformational entropy for the complete ensemble.

24. A computer program according to claim 23 further comprising modeling for each peptide backbone structure of said ensemble in relation to said MHC molecule, at least the side-chains of said peptide.

25. A computer program according to claim 23 or 24 wherein said peptide backbone structures are obtained by computer modeling or by retrieval from a database.

26. A device for evaluating the binding affinity of a peptide for an MHC class I or class II molecule, comprising:

- receiving an ensemble of representations of structures of the complex between said MHC molecule and said peptide,
- evaluating one or more components of the potential energy of each complex of the ensemble,
- evaluating the conformational entropy for the complete ensemble.

27. A peptide which binds MHC class I or class II molecules, said peptide being obtainable by using the methods of any of claims 1 to 21.

28. An peptide which binds MHC class I or class II molecules, said peptide being obtained by using the methods of any of claims 1 to 21.

29. A nucleic acid encoding a peptide as defined in claim 27 or 28.

30. A nucleic acid of at least 15 nucleotides in length specifically hybridizing with the nucleic acid of claim 29.

31. An antibody specifically recognizing a peptide according to claim 27 or 28.

32. An antibody specifically recognizing a nucleic acid according to claim 29 or 30.

33. The peptide according to claim 27 or 28 for use as a medicament.

34. The nucleic acid according to claim 29 or 30 for use as a medicament.